**REMARKS**

Claims 1-6, 11-17, 59-61, 65, 67, 68, 72 and 73 presently appear in this case. Claims 57 and 58 were never a part of this case as they were not entered. The summary page of the official action of June 25, 2008, indicates that claims 57 and 58 were withdrawn from consideration; however, page 2 properly indicates that they are claims that were not entered. No claims have been allowed. The official action of June 25, 2008, has now been carefully studied. Reconsideration and allowance are hereby respectfully urged.

Briefly, the present invention relates to a method of altering gene expression in a population of human embryonic stem cells by introducing into the population of human embryonic stem cells a polynucleotide that contains a gene expression altering sequence. It is possible to obtain a transfection efficiency greater than that obtainable by means of electroporation by use of a transfection reagent that is a linear polymer of polyethyleneimine.

Claims 1-7, 11-17, 59-61, 65-68 and newly added claim 71 have been rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement by adding new matter. The examiner states that reciting that the transfection efficiency is greater than that

obtainable by means of electroporation using a single 625 V/cm pulse at room temperature is new matter.

The claims have now been amended to delete the language that the examiner considers to be new matter, thus obviating this rejection.

Claims 1-7, 11-17, 59-61, 65-68 and 71 have been rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the enablement requirement. The examiner states that the claims as written provide no specific parameter with which to compare the other transfection reagents relative transfection efficiency. This rejection is respectfully traversed.

As indicated above, the claims have been amended to delete reference to the specific transfection method used. Nevertheless, the present specification discloses the preferred electroporation conditions that are used with murine ES cells. Furthermore, it teaches in the same paragraph (page 11, beginning at line 24, states that electroporation was also used with human cells. This is also disclosed in the examples. Those of ordinary skill in the art reading the present specification would have understood that, in the absence of any specification of electroporation conditions in the examples, the electroporation conditions used for the human cells would have been the same as those used for the

murine cells. There was no reason *ab initio* to believe that any modification of conditions would be necessary for human cells. Indeed, the examiner's art rejection of claims 72-74 suggests that there would be no difference in transfection conditions for any animal cells, including murine and human.

In any event, all of the present claims are now limited to the use of a transfection reagent that is a linear polymer of ethyleneimine. The present specification establishes that the transfection efficiency using such a transfection reagent is greater than that obtained using electroporation. It is no longer necessary to specify that the transfection reagent is one that provides a transfection efficiency greater than that obtainable by electroporation using a single 625 V/cm pulse at room temperature. The present claims require no selection of transfection reagent. The present claims are limited to that transfection reagent that has been shown to be unexpectedly superior to electroporation. Those of ordinary skill in the art would know exactly how to use the present invention as one only needs to use a linear polymer of ethyleneimine. One does not have to know electroporation conditions in order to practice the present invention. As there is no selection of transfection reagents being made in accordance with the present claims, all of the present claims should now be free

of the present rejection for the same reason that claim 72-74 were indicated to be free thereof. Reconsideration and withdrawal of this rejection are therefore respectfully urged.

Claims 72-74 have been rejected under 35 U.S.C. 103(a) as being unpatentable over Smith in view of Blanche. The examiner states that Smith teaches the generation of genetically modified stem cells using the term "animal cell" that embraces human cells. The examiner states that Smith teaches that various means of introducing the selectable marker may be employed such as transfection, viral vector, lipofection or electroporation. The examiner concedes that Smith does not teach transfection in the presence of a linear polymer of polyethyleneimine. The examiner states that Blanche teaches various compositions that are useful for nucleic acid transfection techniques and in particular linear polyethyleneimine (PEI). The examiner states that Blanche teaches that PEI has an entirely advantageous property, citing column 5, lines 25-45, and claims 1 and 5. The examiner considers that it would have been obvious to modify the transfection techniques of human ES cells as suggested by Smith and utilize a linear PEI, as taught by Blanche with a reasonable expectation of success. This rejection is respectfully traversed.

It is essentially the examiner's position that the use of PEI is merely one known way to accomplish transfection and therefore it would have been obvious to use it in Smith. However, any *prima facie* case of obviousness established by the examiner has been overcome by the evidence of unexpected results that are present in the specification. Nothing in Smith or Blanche would suggest that the use of PEI would yield transfection efficiencies greater than those obtainable using the best technique that was previously known for transfecting murine ES cells, i.e., electroporation. The present specification has comparative examples of PEI with electroporation showing the unexpected superiority of PEI. It discloses the electroporation conditions used with murine ES cells and, for the purpose of this comparison, it is presumed that the same conditions were used in the comparative experiments in the present specification. It is not necessary for the claim to specify electroporation conditions.

The examiner has not explained why this new rejection is applicable to claims limited to the use of PEI while all of the previous art rejections set forth in the official action of October 4, 2007, were not considered to be applicable to claims limited to PEI, i.e., claims 7 and 66. It is not necessary for the claims to recite that the process yields unexpected efficiency over electroporation. It is

sufficient that the evidence of record establishes unexpected results that rebut the examiner's *prima facie* case of obviousness. The examiner has not applied this rejection to any of claims other than claims 72-74, suggesting that the only way to overcome the rejection is to insert into the claim what electroporation conditions were used in the examples. However, this is not something that needs to be in the claim. The examiner has not stated any reasoning for doubting the evidence of unexpected results of record in this case, i.e., appearing in the present specification. Accordingly, claims 72 and 73 should be unobvious over the prior art cited by the examiner after considering the evidence of unexpected results in the present specification, particularly in light of the fact that the examiner did not apply this rejection to previously appearing claims 7, 66 and 71.

Claim 1 is now identical to claim 72, except specifying that the method is for altering gene expression with a transfection efficiency greater than that obtainable by means of electroporation. However, it is not necessary that these words appear in the claim in order for any *prima facie* case of obviousness to be rebutted. Claim 11 differs from claim 73 in the same manner. Newly amended claim 65 is now identical to claim 4, and thus claim 74 has been deleted.

Accordingly, in view of the evidence of unexpected results (which were convincing to establish the unobviousness of claims 7 and 66 at the time of the official action of October 4, 2007, i.e., prior to the time that the alleged new matter was added to the claims), reconsideration and withdrawal of this rejection are respectfully urged.

It is submitted that all of the claims now present in the case clearly define over the references of record and fully comply with 35 U.S.C. 112. Reconsideration and allowance are therefore earnestly solicited.

Respectfully submitted,

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